



## Physical Anthropology Section - 2013

### H61 Sampling Procedure for the Histological Analysis of Pediatric Skeletal Trauma

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After attending this presentation, attendees will appreciate the use of hard tissue histology in evaluating pediatric trauma and be introduced to a skeletal sampling procedure that forensic anthropologists should consider when using bone histology to examine pediatric remains.

This presentation will impact the forensic science community by demonstrating the growing role of anthropology in the recognition and interpretation of pediatric fractures and the importance of skeletal sampling procedures to maximize evidence obtained during the skeletal survey.

The evaluation of fractures in children is vital for interpreting an injury as non-accidental and in determining if a history of injury exists. Thus, complete documentation of pediatric skeletal injury is paramount in diagnosing abuse.<sup>1</sup> The common anthropological approach to skeletal analysis is to remove the soft tissue and other organic components using a maceration procedure, leaving dry bone for analysis. Love and Sanchez discussed an approach to grossly examine the pediatric skeleton that requires the meticulous reflection of soft tissues at autopsy to expose the osseous and chondral tissues.<sup>1</sup> In addition to this approach, sampling fracture sites or potential fracture sites for histological analysis may identify fractures not seen during the gross examination or provide useful information as to the mechanism and/or timing of the injury that would be otherwise lost due to the maceration procedure. Furthermore, the typical anthropological assessment of fracture healing relies upon the observation of gross bony changes, which will appear secondary to histological changes.

Owing to the difficulty in evaluating pediatric skeletal trauma, a sampling technique was developed to remove specific fractures or segments of fractures for histological analysis as a part of the anthropological exam. The first step is performing a radiological and gross skeletal survey to determine if histological analysis would be beneficial and, if so, select the sampling area(s). The following criteria assist with the sample selection: Sampling is considered for (1) fractures with no gross evidence of healing; (2) fractures that demonstrate differential healing; (3) areas overlying hemorrhage with no apparent bone injury; and, (4) bones suspected of injury that contain chondro-osseous junctions.

Following the gross evaluation and documentation of a selected sampling site, the sample is removed for histological preparation. A small rotary tool with a cut-off wheel is used to remove a segment of the fractured bone. For example, a segment of a skull fracture is removed by cutting a window out of the fracture. An alternate method is to remove the bone in its entirety. Following specimen removal, the tissue is fixed in a 10% formalin solution for 24 hours and submitted to the Histology Laboratory within the Medical Examiner's Office for decalcification. Depending on the specimen, decalcification may take 1 – 3 days.

Once decalcified, the sample can be trimmed using a microtome blade or scalpel in order to fit the sample into a histological cassette for embedding. It is important to leave a portion of "normal" or non-traumatic bone on either side of the fracture so one can examine the lamellar bone leading to the fracture margins or callus, if present. The sample is then placed into the histological cassette and positioned for the desired histological section (e.g., cross section or longitudinal section). The samples are embedded into paraffin blocks and slides are prepared following standard histological procedures. A hematoxylin-eosin stain is applied to assist with the identification of histological features (i.e., new bone formation, mineralized callus, cartilaginous tissue, or fibrotic tissue). Finally, the anthropologist reviews the histological slides with the pathologist to evaluate any microscopic findings.

While this technique requires additional time to perform the analysis of pediatric skeletal trauma, it provides further opportunity to describe fractures that may be difficult to identify or fully characterize at the gross and radiological level. In addition, histological analysis may provide for a more accurate determination of the timing of an injury considering that early reparative response to a bone fracture will not manifest at the gross level until osteoblasts and osteoclasts are given time to respond to the injured tissue. Therefore, the sampling technique described should be considered by anthropologists working with pediatric remains to ensure that skeletal injuries are fully characterized.

#### Reference:

- <sup>1</sup> Love JC, Sanchez LA. Recognition of skeletal fractures in infants: an autopsy technique. *J Forensic Sci* 2009;54(6):1443-6.

#### Pediatric Trauma, Histology, Fracture Analysis